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METHOD AND MEDICAMENT FOR INHIBITING THE EXPRESSION OF A DEFINED GENE

METHODE ET MEDICAMENT DESTINES A INHIBER L'EXPRESSION D'UN GENE DONNE

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- **UHLMANN E ET AL: "ANTISENSE OLIGONUCLEOTIDES: A NEW THERAPEUTIC PRINCIPLE" CHEMICAL REVIEWS,US,AMERICAN CHEMICAL SOCIETY. EASTON, Bd. 90, Nr. 4, 1. Juni 1990 (1990-06-01), Seiten 543-584, XP000141412 ISSN: 0009-2665**
- **MADHUR K. ET AL.: "Antisense RNA : function and fate of duplex RNA in cells of higher eukaryotes." MICROBIOLOGY AND MOLECULAR BIOLOGY REVIEWS, Bd. 62, Dezember 1998 (1998-12), Seiten 1415-1434, XP000909741**

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**EP 1 144 623 B9**

# EP1144623 GRANTED CLAIMS - ENGLISH

International Patent Application No. PCT/DE00/00244  
of Dr Roland Kreutzer and Dr Stefan Limmer

## New Patent Claims

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1. Method for inhibiting the expression of a given target gene in a cell in vitro, where an oligoribonucleotide with double-stranded structure (dsRNA) formed by two separate RNA single strands is introduced into the cell, where one strand of the dsRNA has a region which is complementary to the target gene, characterized in that the complementary region has less than 25 successive nucleotide pairs.

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2. Method according to claim 1, where the dsRNA is enclosed by micellar structures, preferably by liposomes.

3. Method according to either of the preceding claims, where the dsRNA is enclosed by natural viral capsids or by chemically or enzymatically produced artificial capsids or structures derived therefrom.

4. Method according to one of the preceding claims, where the target gene is expressed in eukaryotic cells.

5. Method according to one of the preceding claims, where the target gene is selected from the following group: oncogene, cytokin gene, Id-protein gene, development gene, prion gene.

6. Method according to one of the preceding claims, where the target gene is expressed in pathogenic organisms, preferably in plasmodia.

New Patent Claims

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1. Method for inhibiting the expression of a given target gene in a cell in vitro, where an oligoribonucleotide with double-stranded structure (dsRNA) formed by two separate RNA single strands is introduced into the cell, where one strand of the dsRNA has a region which is complementary to the target gene, characterized in that the complementary region has less than 25 successive nucleotide pairs.

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2. Method according to claim 1, where the dsRNA is enclosed by micellar structures, preferably by liposomes.

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3. Method according to either of the preceding claims, where the dsRNA is enclosed by natural viral capsids or by chemically or enzymatically produced artificial capsids or structures derived therefrom.

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4. Method according to one of the preceding claims, where the target gene is expressed in eukaryotic cells.

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5. Method according to one of the preceding claims, where the target gene is selected from the following group: oncogene, cytokin gene, Id-protein gene, development gene, prion gene.

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6. Method according to one of the preceding claims, where the target gene is expressed in pathogenic organisms, preferably in plasmodia.

7. Method according to one of the preceding claims,  
where the target gene is part of a virus or  
viroid.
- 5 8. Method according to claim 7, where the virus is a  
virus or viroid which is pathogenic for humans.
9. Method according to claim 7, where the virus or  
10 viroid is a virus or viroid which is pathogenic  
for animals or phytopathogenic.
10. Method according to one of the preceding claims,  
where segments of the dsRNA are in double-stranded  
15 form.
11. Method according to one of the preceding claims,  
where the ends of the dsRNA are modified in order  
to counteract degradation in the cell or  
20 dissociation into the single strands.
12. Method according to one of the preceding claims,  
where the cohesion of the double-stranded  
structure, which is caused by the complementary  
25 nucleotide pairs, is increased by at least one,  
preferably two, further chemical linkage(s).
13. Method according to one of the preceding claims,  
where the chemical linkage is formed by a covalent  
30 or ionic bond, a hydrogen bond, hydrophobic  
interactions, preferably van-der-Waals or stacking  
interactions, or by metal-ion coordination.
14. Method according to one of the preceding claims,  
35 where the chemical linkage is generated at at  
least one, preferably both, ends of the double-  
stranded structure.

15. Method according to one of the preceding claims,  
where the chemical linkage is formed by means of  
one or more compound groups, the compound groups  
preferably being poly(oxyphosphinicooxy-  
5 1,3-propanediol) and/or polyethylene glycol  
chains.
16. Method according to one of the preceding claims,  
where the chemical linkage is formed by purine  
10 analogs used in the double-stranded structure in  
place of purines.
17. Method according to one of the preceding claims,  
where the chemical linkage is formed by azabenzene  
15 units introduced into the double-stranded  
structure.
18. Method according to one of the preceding claims,  
where the chemical linkage is formed by branched  
20 nucleotide analogs used in the double-stranded  
structure in place of nucleotides.
19. Method according to one of the preceding claims,  
where at least one of the following groups is used  
25 for generating the chemical linkage: methylene  
blue; bifunctional groups, preferably  
bis(2-chloroethyl)amine; N-acetyl-N'-(p-glyoxyl-  
benzoyl)cystamine; 4-thiouracil; psoralene.
- 30 20. Method according to one of the preceding claims,  
where the chemical linkage is formed by  
thiophosphoryl groups provided at the ends of the  
double-stranded structure.
- 35 21. Method according to one of the preceding claims,  
where the chemical linkage at the ends of the  
double-stranded structure is formed by triple-  
helix bonds.

22. Method according to one of the preceding claims,  
where at least one 2'-hydroxyl group of the  
nucleotides of the dsRNA in the double-stranded  
5 structure is replaced by a chemical group,  
preferably a 2'-amino or a 2'-methyl group.
23. Method according to one of the preceding claims,  
where at least one nucleotide in at least one  
10 strand of the double-stranded structure is a  
locked nucleotide with a sugar ring which is  
chemically modified, preferably by a 2'-O, 4'-C-  
methylene bridge.
- 15 24. Method according to one of the preceding claims,  
where the dsRNA is bound to, associated with or  
surrounded by, at least one viral coat protein  
which originates from a virus, is derived  
therefrom or has been prepared synthetically.
- 20 25. Method according to one of the preceding claims,  
where the coat protein is derived from  
polyomavirus.
- 25 26. Method according to one of the preceding claims,  
where the coat protein contains the polyomavirus  
virus protein 1 (VP1) and/or virus protein 2  
(VP2).
- 30 27. Method according to one of the preceding claims,  
where, when a capsid or capsid-type structure is  
formed from the coat protein, one side faces the  
interior of the capsid or capsid-type structure.
- 35 28. Method according to one of the preceding claims,  
where one strand of the dsRNA is complementary to  
the primary or processed RNA transcript of the  
target gene.



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Anmelder/Applicant/Demandeur/Patentinhaber/Proprietor/Titulaire Ribopharma AG	

# ENTSCHEIDUNG ÜBER DIE ERTEILUNG EINES EUROPÄISCHEN PATENTS GEMÄSS ART. 97(2) EPÜ

Nach Prüfung der europäischen Patentanmeldung Nr. 00910510.7 wird für die benannten Vertragsstaaten ein europäisches Patent mit der Bezeichnung und mit den Unterlagen erteilt, die in der gemäss Regel 51(4) EPÜ ergangenen Mitteilung vom 26.11.01 aufgeführt sind. Den hierzu gegebenenfalls beantragten bzw. vereinbarten Änderungen hat die Prüfungsabteilung zugestimmt. Die vom Anmelder n a c h Eingang der Mitteilung gem. Regel 51(6) EPÜ beantragten und am 21.05.02 beim EPA eingegangenen Berichtigungen wurden berücksichtigt.

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Die Entscheidung wird an dem Tage wirksam, an dem im Europäischen Patentblatt auf die Erteilung hingewiesen worden ist (Art.97(4) und (5) EPÜ).

Der Hinweis über die Erteilung wird im Europäischen Patentblatt 02/35 am 28.08.02 bekanntgemacht.

Prüfungsabteilung  
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HARS J

MERCKLING V



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(54) METHODE ET MEDICAMENT DESTINES A INHIBER L'EXPRESSION D'UN GENE DONNE

(54) METHOD AND MEDICAMENT FOR INHIBITING THE EXPRESSION OF A GIVEN GENE

(57)

<sup>222</sup>The invention relates to a medicament containing at least one double-stranded <sup>2</sup>oligoribonucleotide (dsRNA) designed to inhibit the expression of a target <sup>2</sup>gene. According to the invention, one strand of the dsRNA is at least in part <sup>2</sup>complementary to the target gene.<sup>2</sup>

CANADIAN EQUIVALENT  
OF EP 1144623



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